arrangement thereof. Accordingly, general nucleotides such as a, t, c, g, or u can be used in combination, or alternatively the labeled nucleotide to be used may also be a nucleotide exhibiting weak affinity in base pairing such as inosinic acid as described above. Labeled nucleotides in a nucleotide sequence used for labeling are exemplified by a nucleotide substituted with ^{32}P. Nucleotide derivatives used for labeling is exemplified by nucleotides with a functional group such as a fluorescent compound or ligand having binding capacity. Compounds derivatizing a nucleotide are exemplified by the following compounds:

Fluorescent compound: fluorescein, rhodamine, aminomethyl coumarin; and

Ligand having binding capacity: digoxigenin, biotin.

A kit for preparing hybridization probes can be provided by pre-arranging items necessary for implementing the 3'-tailing labeling method based on the present invention as described herein. The kit of the present invention essentially contains nucleotides and/or nucleotide derivatives, nucleotide compounds, and terminal transferase and, further, can combine buffer preferably used in the reaction, stopping agent for the reaction, reagents to be used for recovering DNA after the reaction, and such.

The hybridization probe of the present invention can be utilized to detect a variety of nucleic acids. Specifically, the probe can be used widely, e.g., for cDNA screening, detection of genes of pathogenic microorganisms and viruses, or analysis for mutation in oncogene. The hybridization probe of the present invention can be used in various assay formats. Specifically, the probe can be used in any of publicly known assay formats, e.g., in dot-blot hybridization assay to observe the reaction of the probe to a target DNA immobilized on a filter, or in *in situ* hybridization assay to observe the localization of nucleic acids on a fixed tissue section. Some assay methods, in which hybridization probes provided by the 3'-tailing labeling method, are already known (Molecular Cloning, Cold spring harbor laboratory press, 1989). However, the occurrence of hybridization due to the nucleotide sequences newly added for labeling is markedly reduced, and therefore masking treatment with

polynucleotide is unnecessary. In addition, the object of hybridization is not limited to DNA, and RNA can also be the object. Further, the probe can be used to screen colonies and plaques.

Gene expression profiling utilizing array technologies is described below as an application example of the inventive hybridization probe based on the high sensitivity and specificity thereof. With the inventive hybridization probe with high sensitivity and high specificity, it is possible to provide accurate expression profiling for a number of RNAs without normalizing arrays. Further, the inventive hybridization probe can be used to detect single nucleotide polymorphisms (SNPs).

Specifically, RNAs, or the cDNAs thereof to be tested for the expression remain unlabeled and they are spotted as mixtures onto a single support to provide an array. The samples to be used can be cDNAs derived from a variety of tissues, or cDNAs from cells collected with time after some drugs are given to the cells. The array is subjected to hybridization treatment with an excess amount of a probe (first probe) of the present invention, which is specific to the gene to be tested. The signal derived from the probe is detected to analyze the level of expression. Subsequently, the resulting double-stranded nucleotide is exposed to denaturation conditions selected based on the Tm value of the probe and thereby the probe is released from the target nucleotide sequence immobilized on the array. The denaturation conditions may be achieved by controlling temperature. There is a known technology to control hybridization electrically (Nat Biotechnol (1999), 17, 365-370). Subsequently, another probe (second probe) is selected for the next gene. Then, the expression level is analyzed in the same manner. This series of treatments is repeated as desired, depending on the number of genes to be tested. A single array can be used repeatedly in this method, and thus expression profiles for many genes can be analyzed with a high degree of precision. The series of treatments can readily be automated. Thus, it can be found that the use of the inventive hybridization probe can rapidly make gene expression profiling efficient. As the matter of course, it is needless to say that the use of the inventive hybridization probe brings similarly high

efficiency to the analyses of SNP.

The principle of the method as described herein is the same as that of the so-called dot-blot assay with cDNA or RNA. However, the method of the present invention can be characterized mainly by using oligo DNA probes with high sensitivity and high specificity and by performing sequential hybridization of multiple distinct probes to a single array. Instead of array on which cDNA or RNA is immobilized, tissue section can be used as a sample (Nature Med. (1998), 4, 844-847) for the implementation of analyses making the most of these advantageous features.

Brief Description of the Drawings

Figure 1 illustrates a photograph showing the results of hybridization assay using an oligonucleotide labeled by 3'-tailing with deoxyinosinic acid as a spacer in accordance with the present invention. In this figure, dIMP indicates deoxyinosinic acid and dAMP indicates deoxyadenylic acid.

Figure 2 illustrates a photograph showing the results of Northern hybridization using an oligonucleotide probe labeled by 3'-tailing with deoxyinosinic acid as a spacer in accordance with the present invention. In this figure, the single band indicates that the probe specifically detected the target mRNA.

Best Mode for Carrying out the Invention

The present invention is described in more detail below with reference to Examples.

Example 1 Labeling of oligonucleotide

(1) An oligonucleotide (ccctacaaagaaaatggagagcct; SEQ ID NO: 1) specific to a gene (accession number: X75861; deposited in the GenBank sequence database administered by National center for biotechnology information (National institutes of health, USA)) was prepared by chemical synthesis (custom-synthesized by GIBCO). The oligonucleotide (100 pmol) was tail-labeled at the 3' end with terminal transferase. The labeling was performed in a solution (0.2 M potassium cacodylate, 25 mM Tris(hydroxy) aminomethane, 0.25 mg/ml bovine serum albumin, 5 mM cobalt chloride, pH6.6) containing 0.5